MAGAGGGLGV WGNLVLLGLC SWTGARAPAP NPGRNLTVET QTTSSISLSW EVPDGLDSQN SNYWVQCTGD GGTTETRNTT 80
ATNVTVDGLG PGSLYTCSVW VEKDGVNSSV GTVTTATAPN PVRNLRVEAQ TNSSIALTWE VPDGPDPQNS TYGVEYTGDG 160
GRAGTRSTAH TNITVDGLEP GCLYAFSMWV GKNGINSSRE TRNATTAHNP VRNLRVEAQT TSSISLSWEV PDGTDPQNST 240
YCIQCTGDGG RTETRNTTDT RVTVDGLGPG SLYTCSVWVE KDGVNSSVEI VTSTTAPNPV RNLTVEAQTN SSIALTWEVP 320
DGPDPQNSTY GVEYTGDGGR AGTRSTAHTN ITVDRLEPGC LYVFSVWVGK NGINSSRETR NATTAPNPVR NLHMETQTNS 400
SIALCWEVPD GPYPQDYTYW VEYTGDGGGT ETRNTTNTSV TAERLEPGTL YTFSVWAEKN GARGSRQNVS ISTVPNAVTS 480
LSKQDWTNST IALRWTAPQG PGQSSYSYWV SWVREGMTDP RTQSTSGTDI TLKELEAGSL YHLTVWAERN EVRGYNSTLT 560
AATAPNEVTD LQNETQTKNS VMLWWKAPGD PHSQLYVYWV QWASKGHPRR GQDPQANWVN QTSRTNETWY KVEALEPGTL 640
YNFTVWAERN DVASSTQSLC ASTYPDTVTI TSCVSTSAGY GVNLIWSCPQ GGYEAFELEV GGQRGSQDRS SCGEAVSVLG 720
LGPARSYPAT ITTIWDGMKV VSHSVVCHTE SAGVIAGAFV GILLFLILVG LLIFFLKRRN KKKQQKPELR DLVFSSPGDI 800
PAEDFADHVR KNERDSNCGF ADEYQQLSLV GHSQSQMV**AS ASENNAKNRY RNVLPYDWSR VPLKPIHEEP GSDYINASFM 880**
PGLWSPQEFI ATQGPLPQTV GDFWRLVWEQ QSHTLVMLTN CMEAGRVKCE HYWPLDSQPC THGHLRVTLV GEEVMENWTV 960
RELLLLQVEE QKTLSVRQFH YQAWPDHGVP SSPDTLLAFW RMLRQWLDQT MEGGPPIVHC SAGVGRTGTL IALDVLLRQL 1040
QSEGLLGPFS FVRKMRESRP LMVQTEAQYV FLHQCILRFL QQSAQAPAEK EVPYEDVENL IYENVAAIQA HKLEV

FIG. 1A

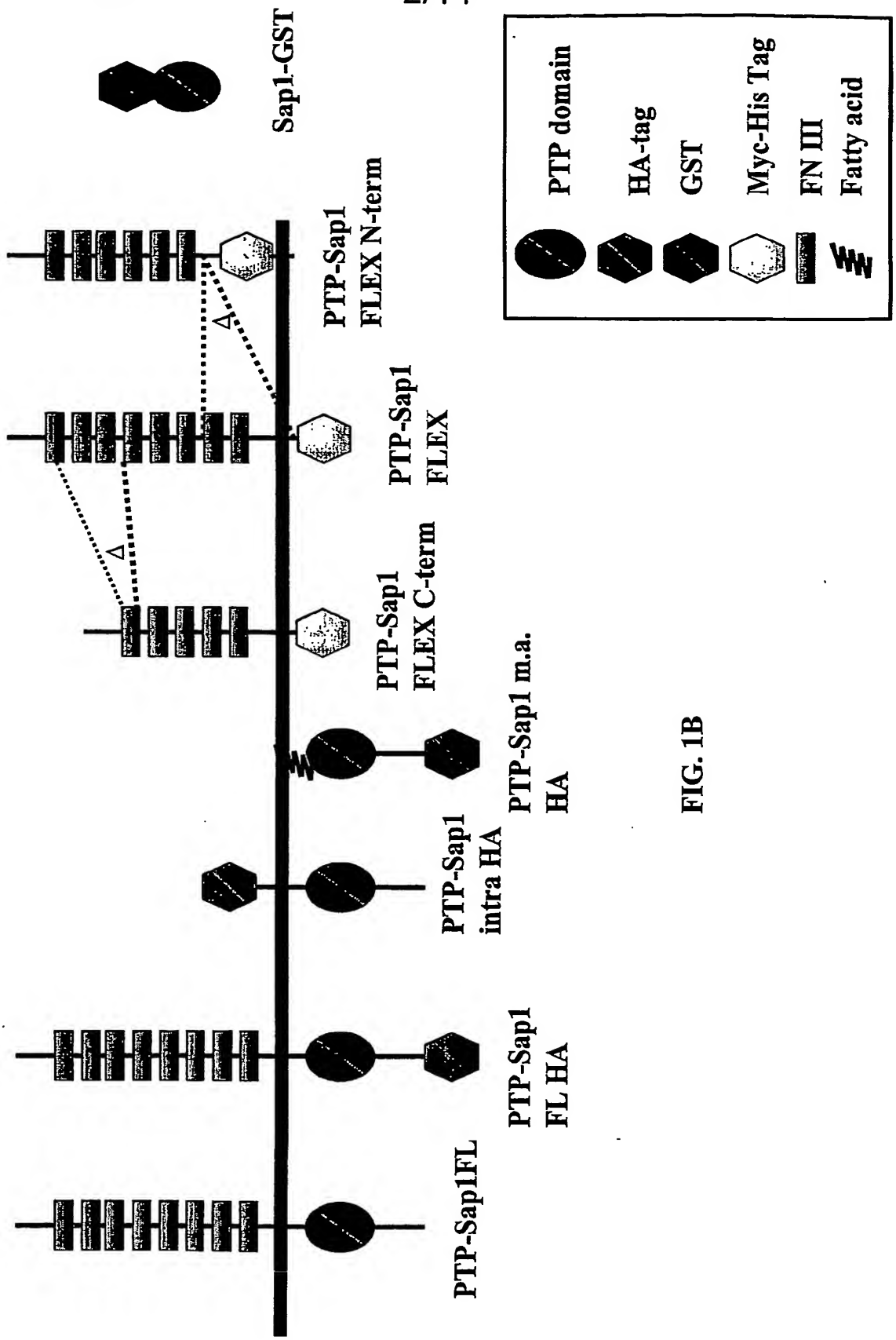
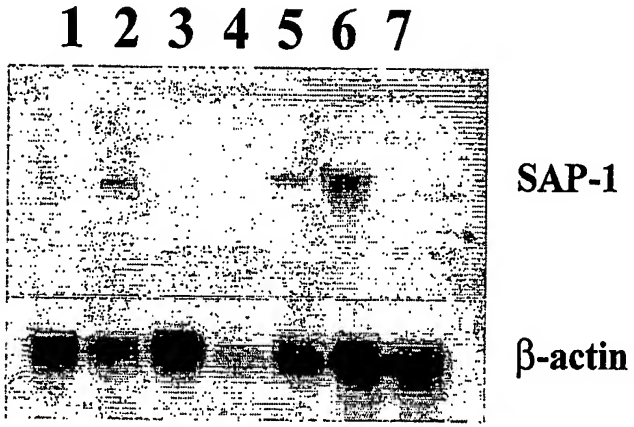

PTP domain
HA-tag
GST
Myc-His Tag
FN III
Fatty acid
Sap1-GST
PTP-Sap1 FLEX N-term
PTP-Sap1 FLEX
PTP-Sap1 FLEX C-term
PTP-Sap1 m.a. HA
PTP-Sap1 intra HA
PTP-Sap1 FL HA
PTP-Sap1FL

FIG. 1B

A



B

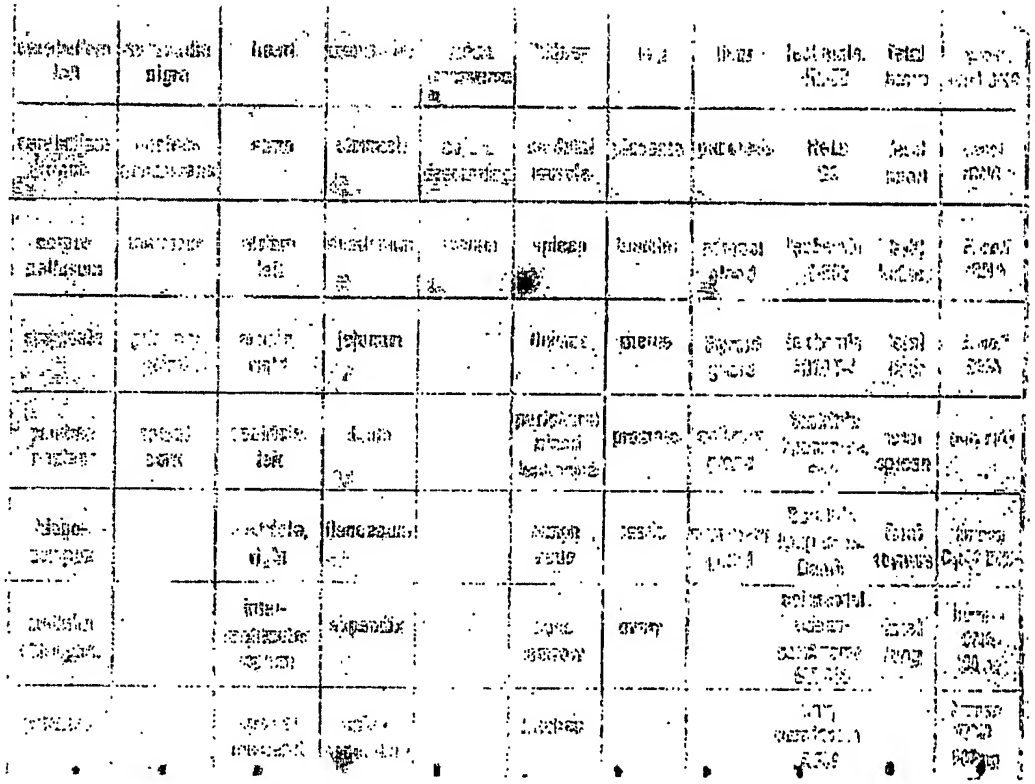


FIG. 2

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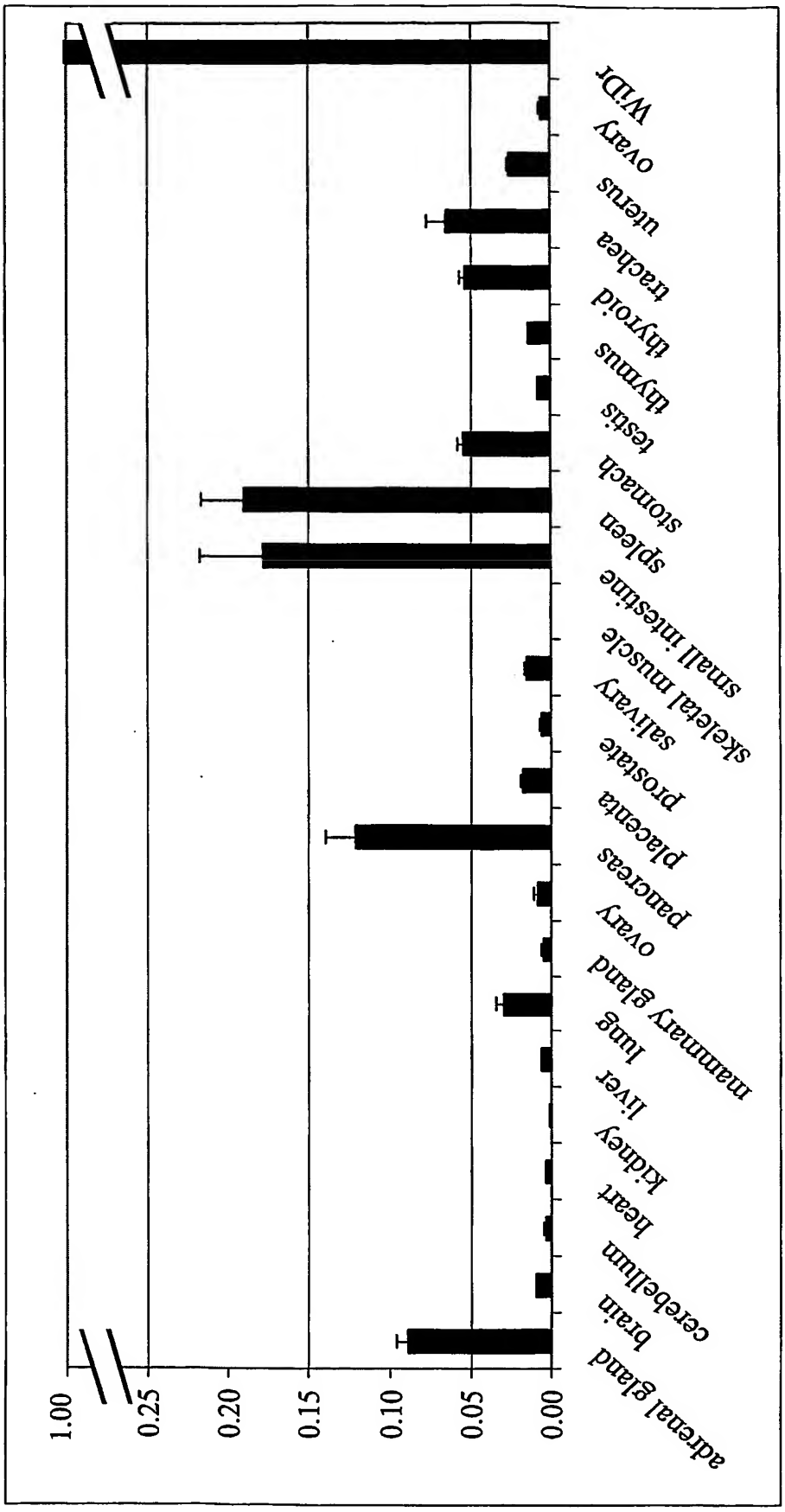

adrenal gland
brain
cerebellum
heart
kidney
liver
lung
mammary gland
ovary
pancreas
placenta
prostate
salivary
skeletal muscle
small intestine
spleen
stomach
testis
thymus
thyroid
trachea
uterus
ovary
WLDr
0.00
0.05
0.10
0.15
0.20
0.25
1.00

Fig. 2 C

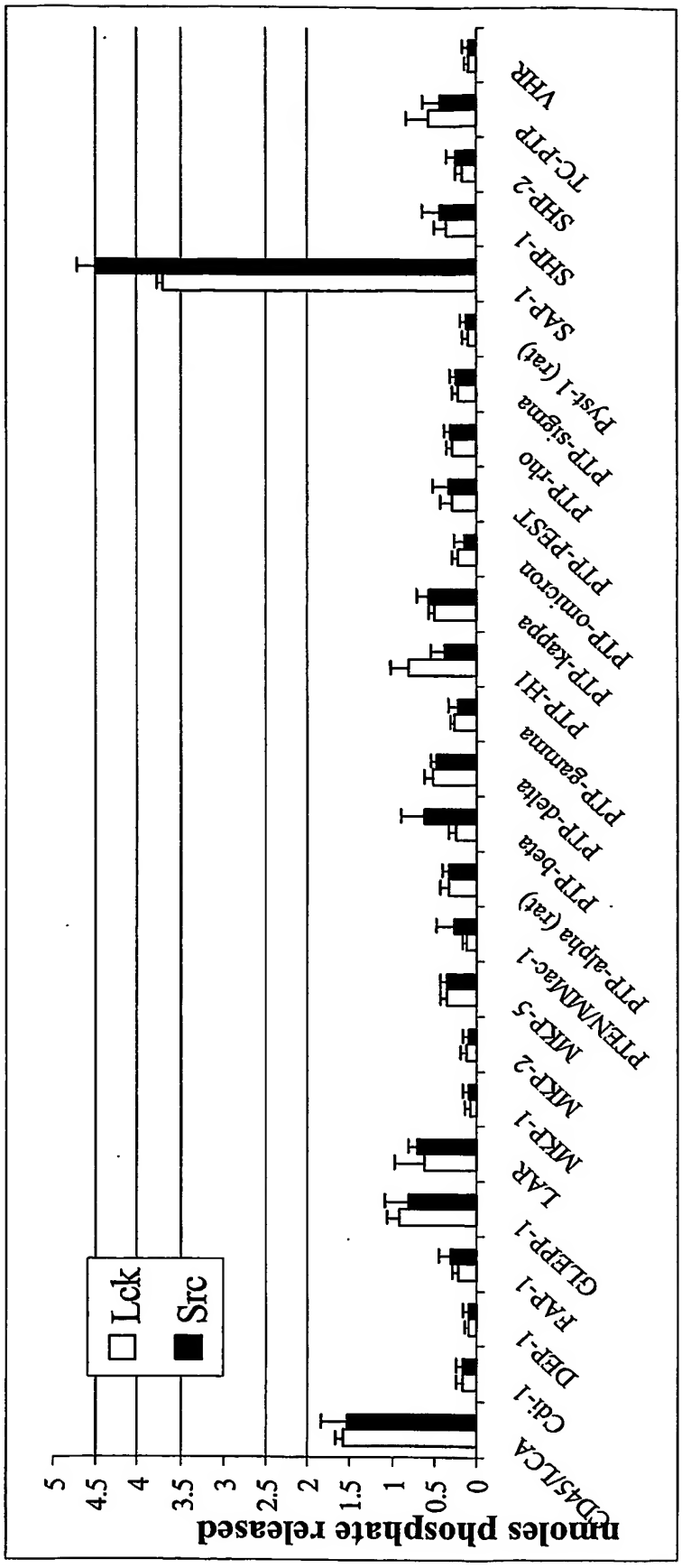

nmoles phosphate released
0
0.5
1
1.5
2
2.5
3
3.5
4
4.5
5
Lck
Src
CD45/LCA
Cdc-1
DEP-1
FAP-1
GLEPP-1
LAR
MKP-1
MKP-2
MKP-5
PTEN/MMac-1
PTP-alpha (rat)
PTP-beta
PTP-delta
PTP-gamma
PTP-H1
PTP-kappa
PTP-omicron
PTP-PEST
PTP-tho
PTP-sigma
Pyst-1 (rat)
SAP-1
SHP-1
SHP-2
TC-PTP
VHR

Fig. 3

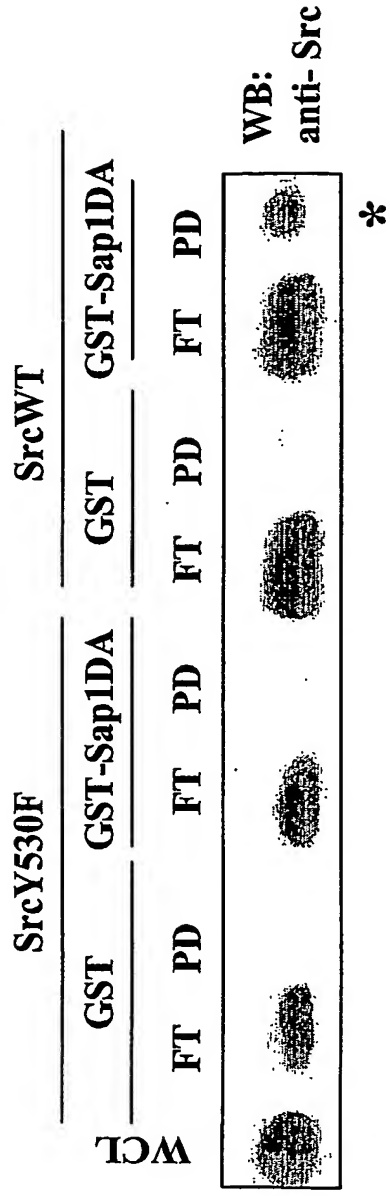

WB:
anti- Src
*
WCL
SrcY530F
SrcWT
GST
GST-Sap1DA
GST
GST-Sap1DA
FT
PD
FT
PD
FT
PD
FT
PD

Fig. 4 A

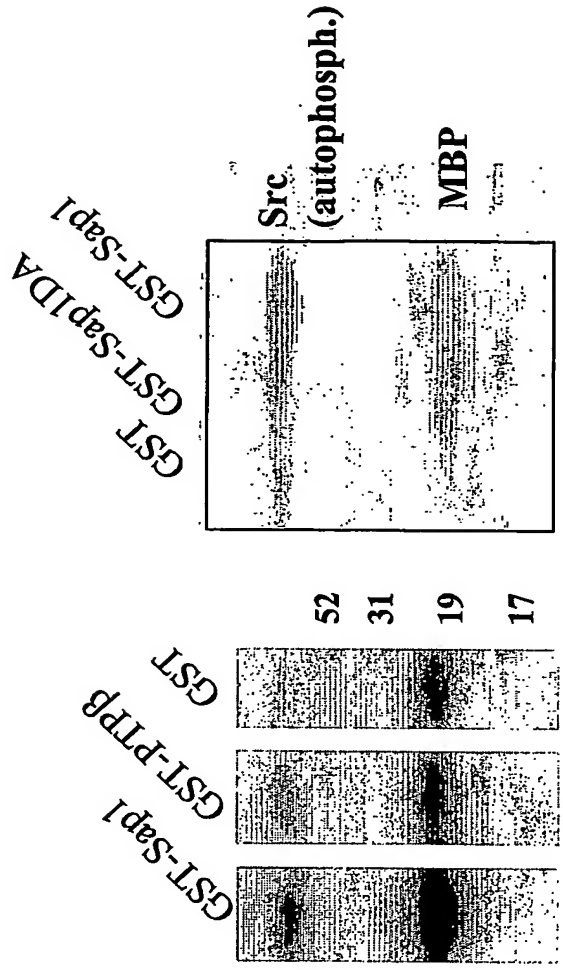

GST
GST-Sap1DA
GST-Sap1
Src
(autophosph.)
MBP
52
31
19
17
GST
GST-PTPβ
GST-Sap1

Fig. 4 C

- GST
+ GST-SAP1
IB: anti Src327
IB: anti TyrP
IP anti-Src327

Fig. 4 B

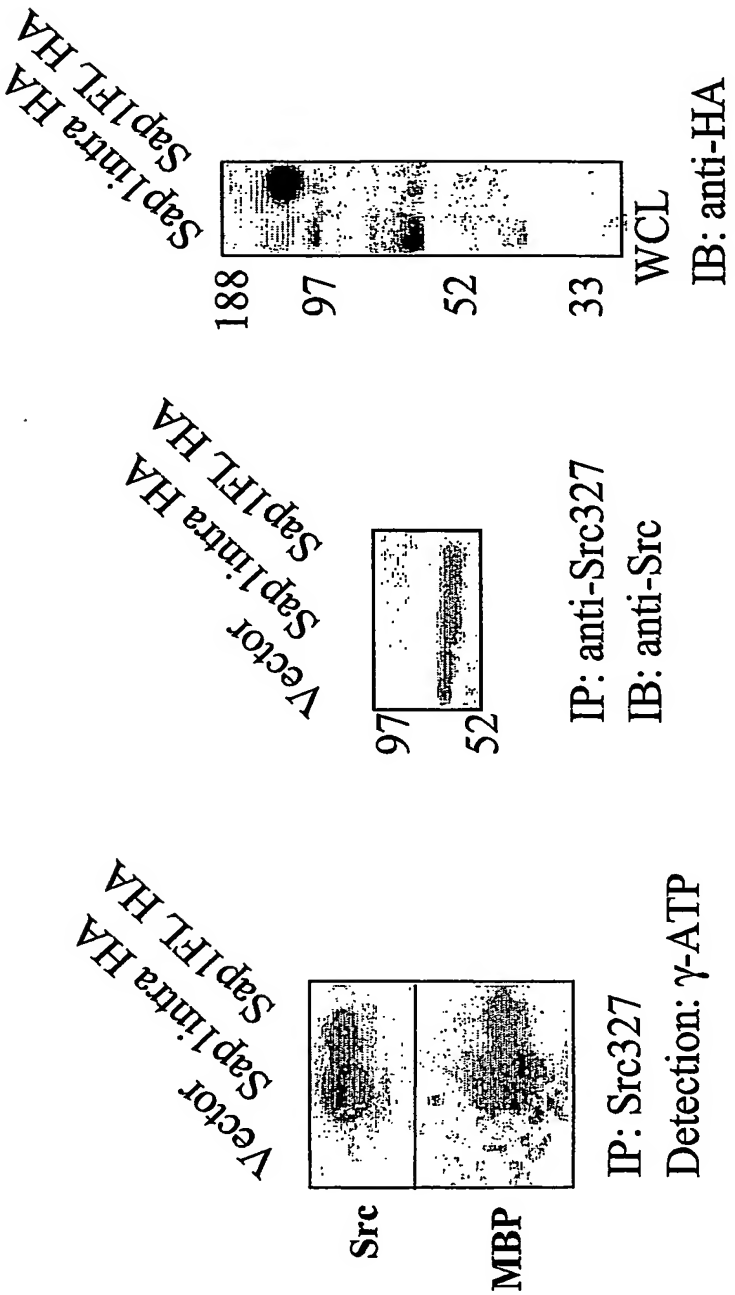

Vector
Sap1intra HA
Sap1FL HA
Src
MBP
IP: Src327
Detection: γ-ATP
Vector
Sap1intra HA
Sap1FL HA
97
52
IP: anti-Src327
IB: anti-Src
Sap1intra HA
Sap1FL HA
188
97
52
33
WCL
IB: anti-HA

Fig. 4 D

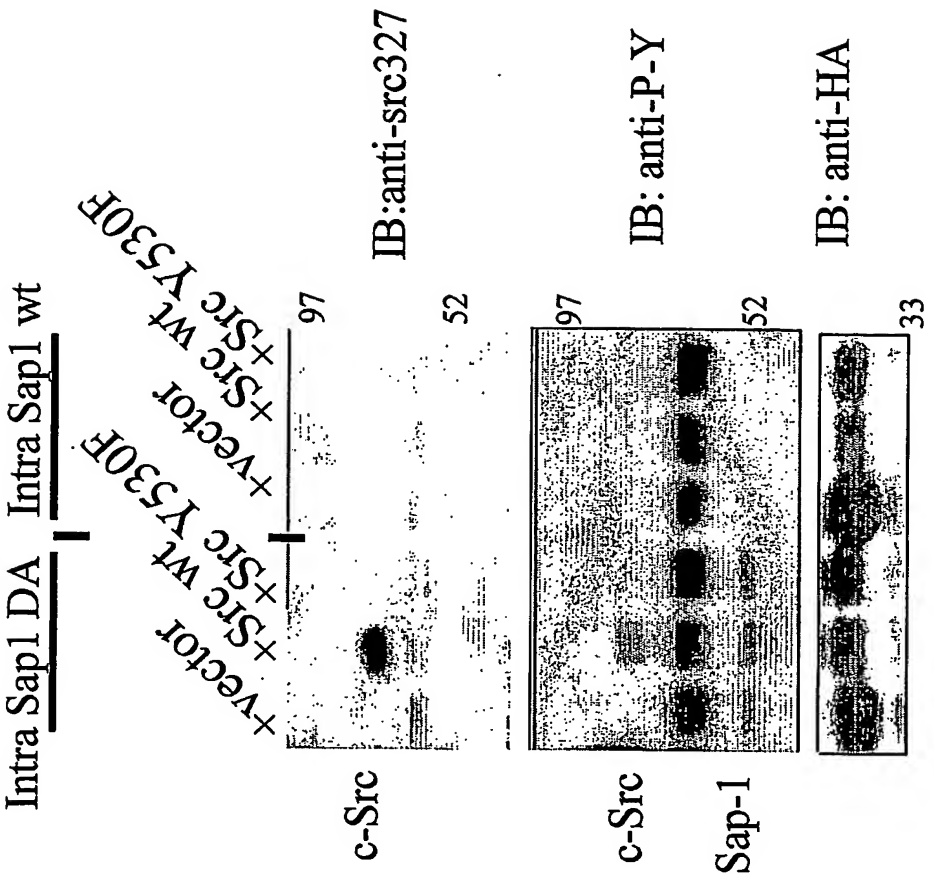

Intra Sap1 DA
Intra Sap1 wt
×Vector
×Src wt
×Src Y530F
×Vector
×Src wt
×Src Y530F
IB:anti-src327
IB: anti-P-Y
IB: anti-HA
97
52
97
52
33
c-Src
c-Src
Sap-1
IP: anti-HA

Fig. 4 F

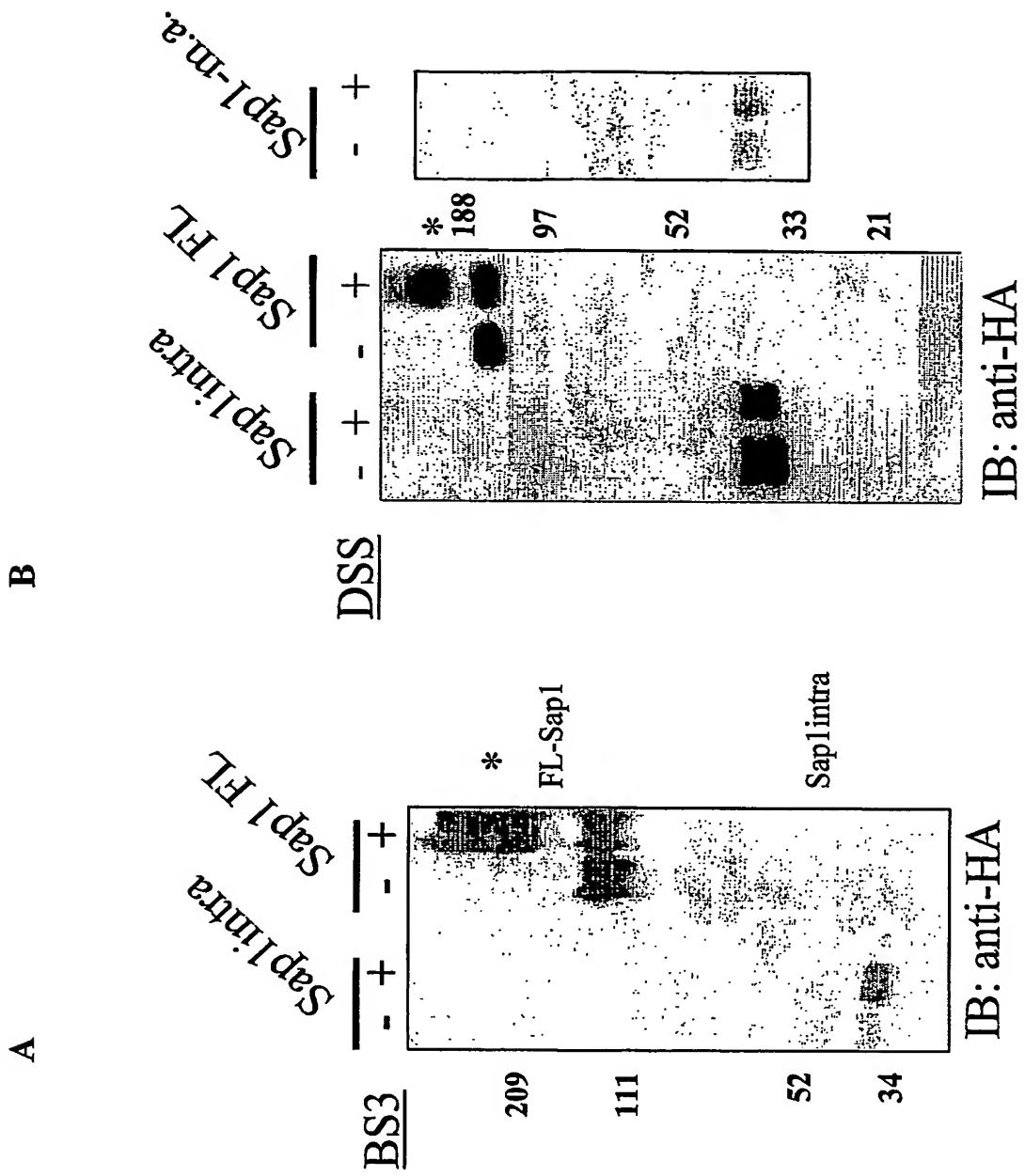

A
BS3
Sap1intra
Sap1 FL
- + - +
209
111
52
34
*
FL-Sap1
Sap1intra
IB: anti-HA
B
DSS
Sap1intra
Sap1 FL
Sap1-m.a.
- + - + - +
*
188
97
52
33
21
IB: anti-HA

FIG. 5

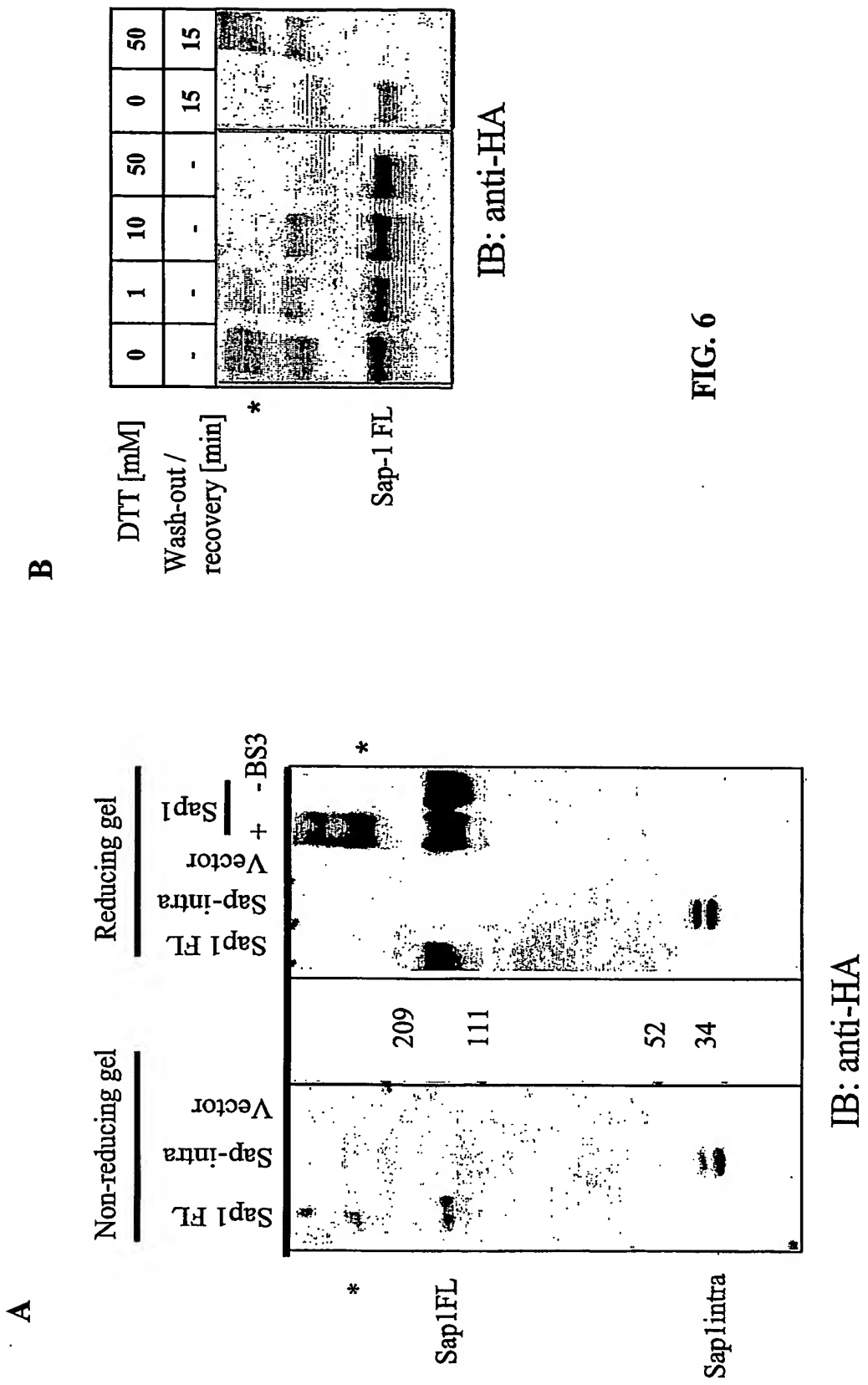

A
Non-reducing gel
Reducing gel
Sap1 FL
Sap-intra
Vector
Sap1 FL
Sap-intra
Vector
+ Sap1
- BS3
209
111
52
34
*
*
Sap1FL
Sap1intra
IB: anti-HA
B
DTT [mM]
Wash-out / recovery [min]
0
1
10
50
0
50
-
-
-
-
15
15
*
Sap-1 FL
IB: anti-HA

FIG. 6

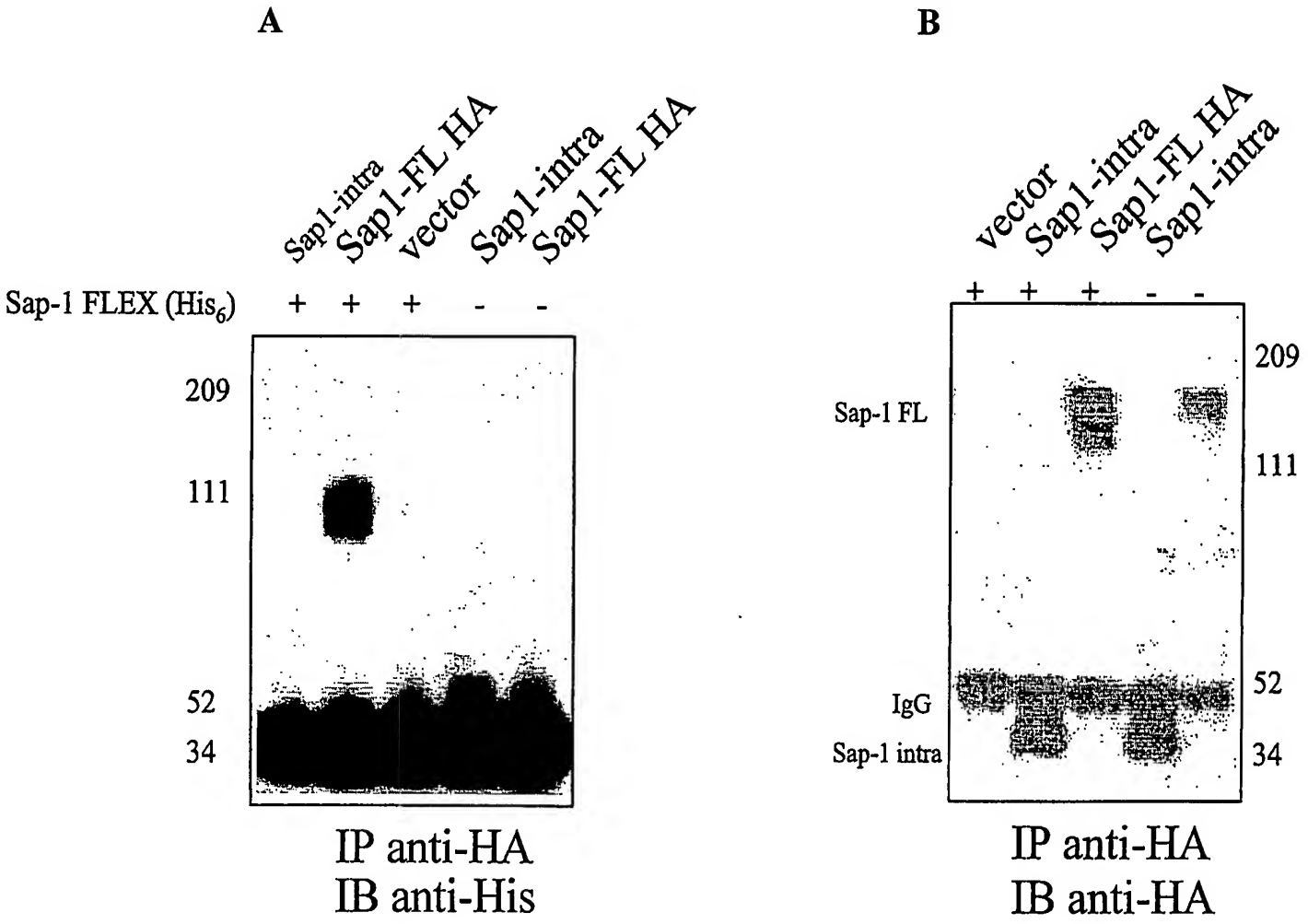

A
Sap1-intra
Sap1-FL HA
vector
Sap1-intra
Sap1-FL HA
Sap-1 FLEX (His6)
+ + + - -
209
111
52
34
IP anti-HA
IB anti-His
B
vector
Sap1-intra
Sap1-FL HA
Sap1-intra
+ + + - -
Sap-1 FL
IgG
Sap-1 intra
209
111
52
34
IP anti-HA
IB anti-HA

FIG. 7

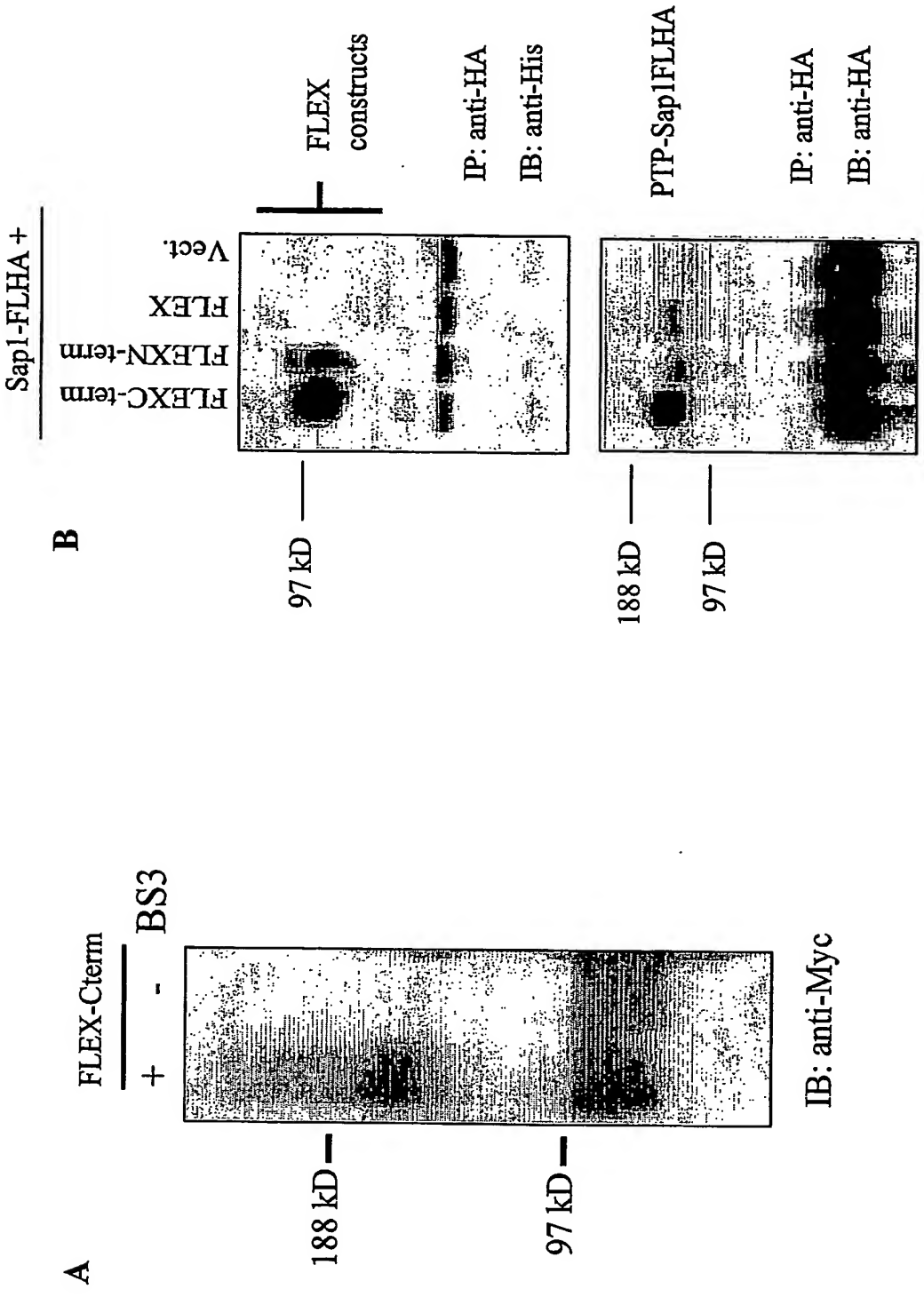
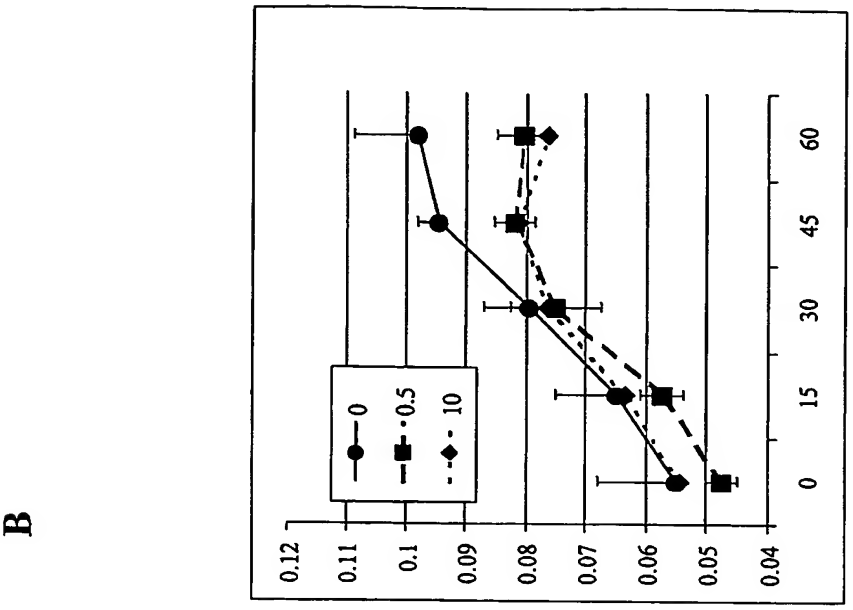

A
FLEX-Cterm
+
-
BS3
188 kD
97 kD
IB: anti-Myc
B
Sap1-FLHA +
FLEXC-term
FLEXN-term
FLEX
Vect.
FLEX constructs
IP: anti-HA
IB: anti-His
97 kD
PTP-Sap1FLHA
IP: anti-HA
IB: anti-HA
188 kD
97 kD

FIG. 8


B
0.12
0.11
0.1
0.09
0.08
0.07
0.06
0.05
0.04
0
15
30
45
60
0
0.5
10

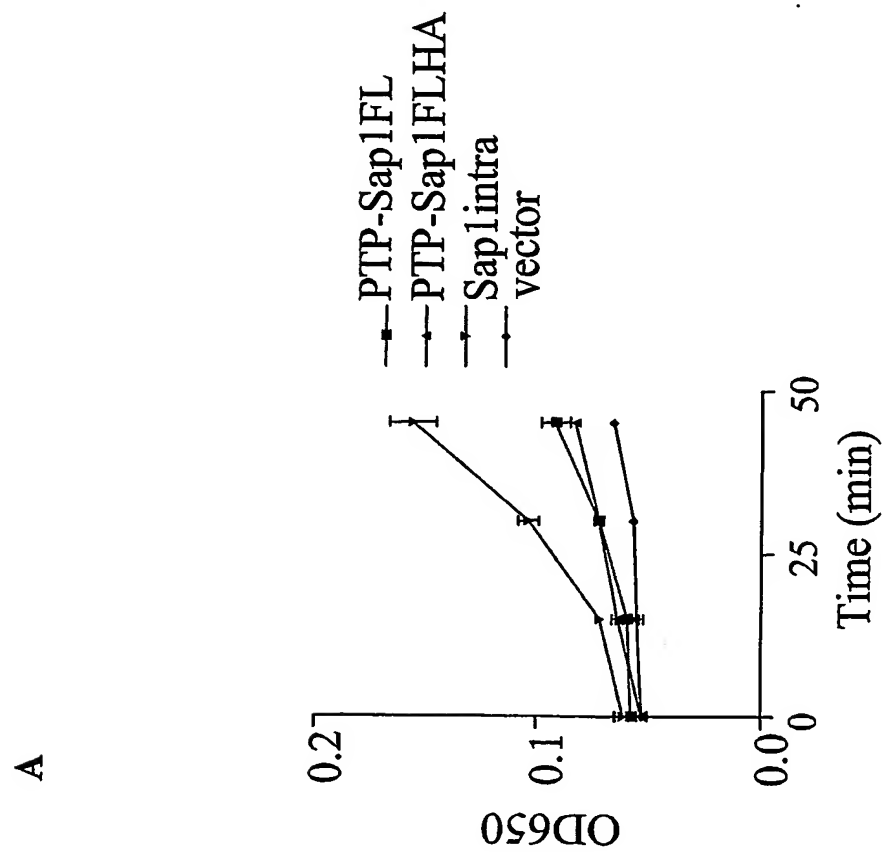

A
PTP-Sap1FL
PTP-Sap1FLHA
Sap1intra
vector
OD650
0.2
0.1
0.0
0
25
50
Time (min)

FIG. 9